Influence of different post-printing treatments on the in-vitro bio-compatibility of three-dimensional generated titanium plates

Ute Ulrike Botzenhart[1], Jakob Schneider[2,3], Elena López[2], Anne Bernhardt[4]

[1]Department of Orthodontics, Medical Faculty Carl Gustav Carus, TU Dresden, 01307 Dresden , Germany

[2]Fraunhofer Institute for Material and Beam Technology, 01277 Dresden, Germany

[3]TU Dresden, 01069 Dresden, Germany

[4]TU Dresden, Centre for Translational Bone, Joint and Soft Tissue Research, 01307 Dresden, Germany

Background

Patient-individual bone plates from titanium alloy (TiAl4V ELI) can be generated by 3D printing. Directly after printing, the samples have a very rough surface requiring further processing.

***The aim of the present study** was to analyze the effect of different grinding and polishing procedures on sample surfaces and in vitro biocompatibility to make sure that the clinically approved material properties of the titanium alloy were not compromised by these treatments.*

Material und Methods

Initial average surface roughness of 3D printed titanium discs (Ti6Al4V ELI) (d=5mm or 10 mm, h=2 mm), produced by selective laser melting (SLM; Ra=22.7 µm), was reduced by sandblasting followed by barrel finishing, electro-polishing, or plasma-polishing and was then evaluated using tactile surface quality measurement (DIN ISO EN 4288). Biocompatibility of the different sample groups (n=6-9 each) was assessed by quantification of metal-ion release, indirect cell viability, and cytotoxicity tests as well as direct cell adhesion, analyzed by fluorescence and scanning electron microscopy (SEM). Mouse fibroblasts (L929), osteosarcoma cells (Saos-2), human primary gingival fibroblast (HPGF) as well as human gingival epithelial cells (HPGEC) were used in this study, respectively. Statistical evaluation was performed with the Kruskal Wallis test, followed by a posthoc Dunn´s test to perform multiple comparisons.

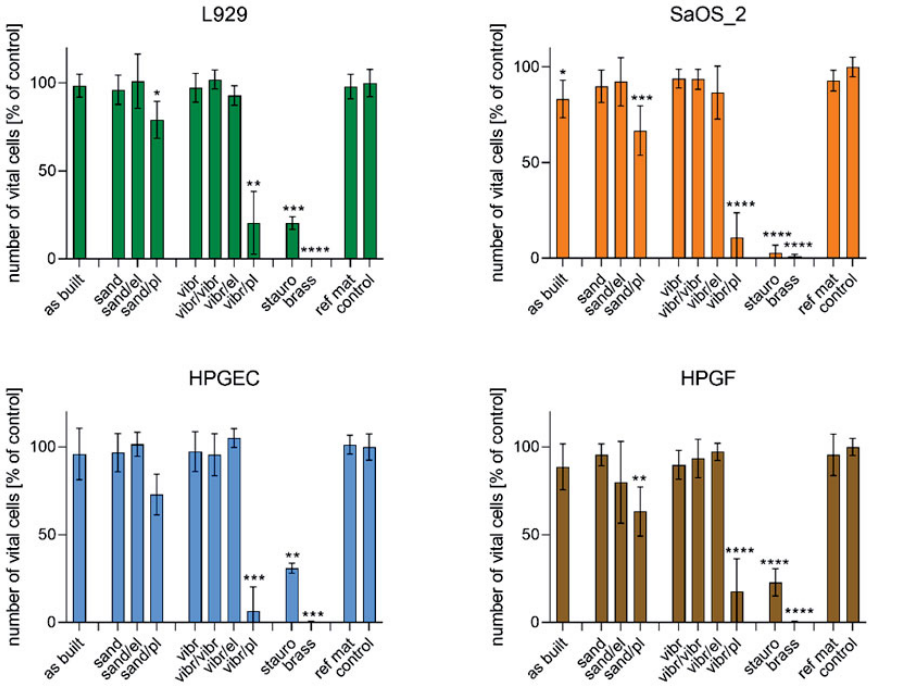

Figure 1:
Indirect cytotoxicity assay. Number of vital cells after cultivating them for 24h in the presence of extracts from different post-treated Ti-6Al-4V samples (sand=sandblasted, el=electro polished, vibr= vibratory polished, pl=plasma polished) compared to the reference material (ref mat=TiAl4V6 by KLS Martin). Number of viable cells was related to the positive control (medium only), which was set to 100 % and negative controls (extracts of brass and 100 µM stauro=staurosporine). Three individual experiments with n = 3 each, average +/- standard deviation, statistical differences were labeled as *($p < 0.5$), **($p < 0.01$) ***($p < 0.001$) and ****($p < 0.0001$).

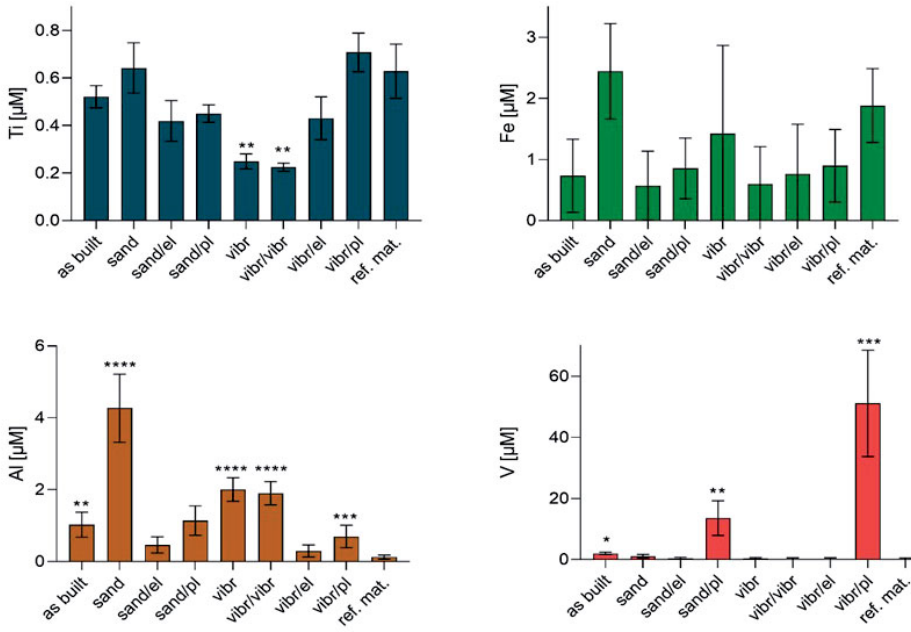

Figure 2:
Concentrations of titanium (Ti), iron (Fe), aluminium (Al) and vanadium (V) ions in extracts of the differently post-treated printed TiAl4V6 samples as described in fig 1. Three individual experiments with n = 3 each, average +/- standard deviation, statistical differences were labeled as *($p < 0.5$), **($p < 0.01$) ***($p < 0.001$) and ****($p < 0.0001$).

Results

All tested treatments were suitable to obtain surface roughness values within the effective roughness spectrum (Ra 0.2 2 µm). Sandblasted, barrel finished, and electro-polished samples showed high cell viability in indirect tests **(Fig. 1)**, as well as good cell adhesion and proliferation when seeded directly with cells. In contrast, plasma-polished samples showed significantly reduced cell viability in indirect tests and very low attached cell numbers after direct colonization, probably caused by the high amount of vanadium ions, which could be found in the cell culture medium after metal-ion release from the plasma-polished samples **(Fig. 2)**.

Conclusion

Bone plates from titanium alloy generated by 3D printing are suitable for clinical use and are in line with the requirements for medical devices according to ISO 10993-5. However, relating to metal-ion release, post-printing treatment must be taken into consideration, critically.

Literature

Bernhardt A, Schneider J, Schroeder A, Papadopoulos K, Lopez E, Brückner F, Botzenhart U. Surface conditioning of additively manufactured titanium implants affects materials properties and in vitro biocompatibility. Mater Sci Eng C Mater Biol Appl. 2021 Feb;119:111631. doi: 10.1016/j.msec.2020.111631. Epub 2020 Oct 15.

The authors would like to thank the Federal Ministry of Education and Research (BMBF) for providing financial support to this project.